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MODIFICATION OF THE REFLEX RESPONSE TO STIMULATION OF CAROTID SINUS BARORECEPTORS DURING AND FOLLOWING STIMULATION OF THE HYPOTHALAMIC DEFENCE AREA IN THE CAT

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SUMMARY

1. The effects of stimulation of the hypothalamic defence area on carotid sinus baroreceptor reflexes have been investigated by examining the cardiovascular responses to a 15 sec period of increased pressure within the vascularly isolated carotid sinus before, during, and immediately following a 25 sec period of hypothalamic stimulation.

2. Identification of the hypothalamic defence area was based on the occurrence of atropine-sensitive muscle vasodilatation. Electrode positions were confirmed by histological examination.

3. During hypothalamic stimulation the reflex fall in blood pressure resulting from a rise in sinus pressure was found to be undiminished whether sinus pressure was raised at the onset or at the 10th sec of hypothalamic stimulation.

4. By contrast, in at least half the cats in which a reflex bradycardia could be evoked from the sinus, this bradycardia was largely if not completely suppressed during hypothalamic stimulation. This suppression of reflex bradycardia occurred when sinus pressure was raised at the onset as well as at the 10th sec of stimulation.

5. During the first 5 sec after hypothalamic stimulation the hypotensive response to an increase in carotid sinus pressure was much reduced; on the other hand the reduction in heart rate was exaggerated, sometimes to a very marked degree.

6. The results suggest that stimulation of the hypothalamic defence area can modify baroreceptor reflexes and that this modification can include selective alterations in the various components of the reflex response.

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INTRODUCTION

While stimulation of localized areas of the brain has been shown by many authors to exert profound effects on the cardiovascular system (see Uvnäs, 1960), less is known about the extent to which such effects may be the results of modification of homoeostatic cardiovascular reflexes. A part of the brain which seems particularly likely to influence baroreceptor reflexes is the region integrating the preparatory stage of the defence reaction. Stimulation of this region, shown by Abrahams, Hilton & Zbrozyna (1960) to lie in the hypothalamus and central grey matter of the mid-brain, produces a pattern of cardiovascular changes, including rises in heart rate and blood pressure, which can be regarded as preparatory for sudden exertion. However, the rise in mean systemic pressure will lead to increased discharge from the arterial baroreceptors. An increased pulse pressure is generally present as well as a rise in mean pressure and this will further enhance the baroreceptor discharge (Ead, Green & Neil, 1952). The normal reflex responses to baroreceptor stimulation, bradycardia and dilatation of the resistance vessels would directly oppose the tachycardia and increased systemic pressure initiated from the defence area and this has led to the suggestion that these baroreceptor reflex effects may be suppressed during the defence response (Hilton, 1963). The present experiments were designed to examine this possibility by quantitatively comparing the changes in heart rate and arterial blood pressure reflexly evoked from the baroreceptors of the vascularly isolated carotid sinus before, during, and immediately following electrical stimulation of sites in the hypothalamus lying within the area integrating the preparatory cardiovascular changes of the defence response. The results indicate that this region can modify the baroreceptor reflex but that different effects are exerted on the various components of the reflex response.

METHODS

Twenty-four cats were used, all but one being female. Their weights ranged from 2·2 to 4·0 kg. Eleven animals were anaesthetized by the intraperitoneal injection of chloralose, 60–70 mg/kg (α-chloralose, Établissements Kuhlmann, Paris, 6–7 ml./kg of a 1 % solution in 0·9 % NaCl) and pentobarbitone, 2 mg/kg (Nembutal, Abbott Laboratories, 60 mg/ml.). Anaesthesia was induced in the remaining animals by inhalation of ether followed by intravenous injection of chloralose, 60–70 mg/kg and pentobarbitone, 2 mg/kg (nine cats), chloralose, 70 mg/kg, by itself (one cat), a mixture of allobarbitone, 50 mg/kg (Dial, Gerhardt-Penick, Ltd.), pentobarbitone, 2 mg/kg and chloralose, 20 mg/kg (two cats), and allobarbitone, 50 mg/kg plus pentobarbitone, 2 mg/kg (one cat). No differences were observed in the responses which could be attributed to the type of anaesthesia and therefore no attempt has been made to subdivide the results on this basis.

Cannulae were inserted into the trachea, a femoral artery and a femoral vein. The femoral artery catheter was advanced into the abdominal aorta for the registra-

tion of arterial blood pressure. A saline–dextrose solution (1 part of 0·9 % NaCl to 4 parts of 5 % dextrose) was infused through the venous cannula at a rate of approximately 6 ml./hr.

Isolation of the carotid sinus. One carotid sinus, usually the right, was prepared for isolation by ligating all the arterial branches arising from the region of the bifurcation with the exception of the external and common carotid arteries. Complete vascular isolation could then be achieved by placing clips on these vessels. Between tests, the clips were removed to restore both the normal pulsatile blood flow to the isolated carotid sinus and the carotid contribution to the cerebral circulation. Care was taken to identify and preserve the sinus nerve, the venous drainage from the carotid body and the nerve branch from the superior cervical ganglion to the sinus region. The contralateral sinus nerve was cut to minimize compensatory changes in the activity of the baroreceptors of the opposite sinus and loops were placed around both aortic nerves so that these could be rapidly divided if necessary.

Following heparinization at a later stage of the preparation (see below) a T-cannula was placed in the common carotid artery. The other arm of the T-cannula led to a simple perfusion apparatus comprising a small reservoir containing saline with compressed air above which enabled non-pulsatile pressures to be applied to the isolated carotid sinus. The compressed air was supplied from two tanks at preset pressures and by turning a tap a stepwise pressure change could be rapidly produced. A side tube from the inlet to the cat allowed the pressure of the perfusate, and hence within the carotid sinus, to be monitored. Completeness of the vascular isolation of the sinus could be judged by the very small volume of saline which entered the cat during the periods of perfusion. Generally this was less than 1 ml. over the course of the entire day. Very often the inflow during a test was insufficient even to wash back all the blood which had crept into the side-arm of the T-cannula during the preceding period of pulsatile blood flow. Since the cannula was inserted into the common carotid artery 2–3 cm below the carotid sinus it is likely therefore that the sinus remained filled with blood throughout each test.

Muscle vascular resistance. Muscle vascular resistance was monitored to confirm activation of the sympathetic vasodilator system which was regarded as an essential criterion for activation of the hypothalamic defence centre. One hind limb was skinned and blood flow to the paw excluded by tying a tight ligature around the ankle. The femoral artery was isolated and its circumflex and other anastomotic branches ligated. After heparinization (see below) the femoral artery was cannulated proximally and distally and the limb circulation, now almost wholly restricted to muscle, maintained at constant volume inflow with a roller pump (H.R. Flow-Inducer, Watson–Marlow, Ltd.). The inflow pressure was measured and the pump speed set to give a perfusion pressure approximately equal to the control systemic pressure.

Stimulation of the hypothalamus. Once the carotid sinus had been isolated and the hind limb prepared for perfusion the cat was placed in the stereotaxic frame (La Précision Cinématographique, Asnières, Seine). The skull was opened and after an interval of 20–30 min to ensure adequate clotting, heparin 1000 i.u./kg (Pularin, Evans Medical, 1000 i.u./ml.) was given, the T-cannula placed in the common carotid artery, the femoral cannulation carried out and the limb perfusion established. The animal was then replaced in the stereotaxic frame and the stimulating electrode advanced into the hypothalamus. The initial placement was carried out with the aid of a stereotaxic atlas (Snider & Niemer, 1961). The final position was determined by testing the response to 5–10 sec periods of stimulation, particular attention being paid to the presence or absence of muscle vasodilatation. In the final stages the electrode was advanced in steps of 0·1 mm.

The stimulating electrode was a ground and electrolytically thinned stainless-steel needle which had been insulated with Araldite 985E (C.I.B.A.) except at the tip; the length of the bared tip was about 50 μ. A chlorided silver plate approximately 1 × 1 cm buried in the subcutaneous tissues of the back of the neck formed the indifferent electrode. Accurately balanced biphasic square-wave pulses were delivered from a pair of isolated stimulators (Devices, Mk IV) triggered by a twin-pulse generator. The stimulus parameters, a frequency of 80 Hz, and a pulse duration of 2 msec, were those reported by Lindgren (1955) as being optimal for the production of a vasodilator response. To minimize tissue damage the stimulus intensity was kept as low as was consistent with an adequate physiological response. Stimulus current was continuously monitored and was generally 100–200 μA, though towards the end of the experiment the stimulus current was sometimes increased up to 300–400 μA. To detect any unidirectional d.c. component a d.c. microammeter was included in the output circuit as an additional monitor. Continuous monitoring of the current provided a useful guide to conditions at the electrode tip; damage to the tip, local tissue destruction or haemorrhage at the site of stimulation being associated with an inconstant current.

Recordings. The systemic arterial pressure, carotid sinus pressure and limb perfusion pressure were measured by transducers (Consolidated Electrodynamics, Type 4-327-L221) and after amplification by carrier amplifiers (S.E. Laboratories, Type 423/1) the signals were displayed on a direct-writing ultra-violet light recorder (S.E. Laboratories, Model No. 2100). Mean arterial pressure was obtained by passing the systemic pressure signal through a simple R–C network with a time constant of 1 sec and was recorded by a separate galvanometer. Another galvanometer gave a record of stimulus current size. Heart rates were measured in the fifteen experiments in which the paper speed was sufficiently fast for the pulsations of the blood pressure record to be counted.

Histological confirmation of electrode position. At the end of the experiment the cat was killed by bleeding and the brain fixed with the electrode *in situ* by perfusing formal-sucrose through the headward limb of the common carotid T-cannula. Sections 10 μ thick were cut and stained with luxol fast blue and cresyl violet (Klüver & Barrera, 1953).

RESULTS

Effects of hypothalamic stimulation

The principal features of the cardiovascular response to hypothalamic stimulation are illustrated in Fig. 1. Stimulation lasted for 30 sec in this instance but in all the experiments referred to subsequently a 25 sec period of stimulation was used. The innervated carotid sinus was not excluded from the rest of the circulation during this test, so that the pressure changes within it paralleled those in the aorta. With the onset of stimulation, muscle perfusion pressure rose briefly, then fell to considerably below its control level, indicating the occurrence of vasodilatation in the isolated hind-limb muscle bed which is characteristic of activation of the hypothalamic defence area. Typically, the vasodilatation did not commence until about 5 sec after the onset of stimulation, and had begun to wane by the 15th sec even though hypothalamic stimulation was still continuing. Systemic arterial blood pressure rose sharply within the first

5 sec of the test from an initial level of 115 mm Hg to a peak of 168 mm Hg, the levels thereafter reflecting to some extent the changes in resistance in the muscle vessels. An initial marked increase was the most commonly observed pattern of systemic pressure change, though in several cats the rise was more gradual and in others there was only a minimal B.P. change. In the experiment shown in Fig. 1 the rise in arterial pressure was accompanied by an increase in heart rate which rose from 276 beats/min in the control period to a maximum of 294 beats/min during stimulation.

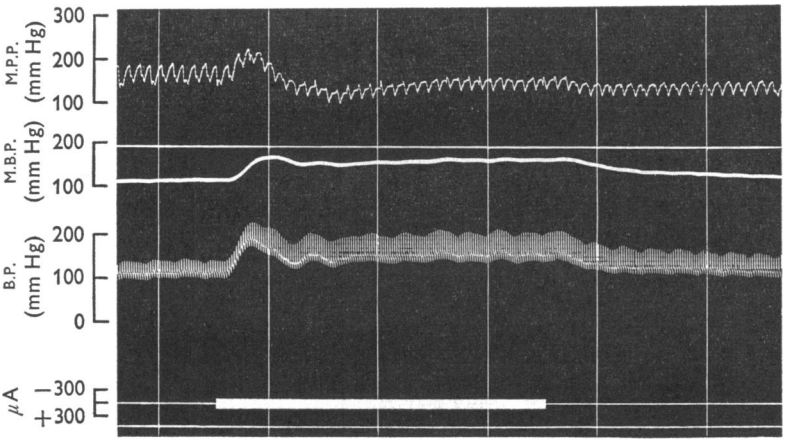

Fig. 1. Changes in mean (M.B.P.) and pulsatile (B.P.) aortic pressure and muscle perfusion pressure (M.P.P.) during a 30 sec period of hypothalamic stimulation. Muscle perfusion pressure refers to the inflow pressure to the skinned hind limb perfused at constant flow. In this and subsequent Figs. the stimulus marker shows the size and direction of the current pulses as well as the duration of the stimulus. One sinus nerve was cut; the other carotid sinus was not excluded from the circulation. Time marker at 10 sec intervals.

The mean changes in systemic blood pressure, heart rate and muscle perfusion pressure during twenty-six stimulations in the fifteen cats in which heart rate was measured are summarized in Fig. 2 which also emphasizes the occurrence of muscle vasodilatation and its transient nature. In these tests the pressure in the innervated, isolated carotid sinus was held constant at pressures within the range 75–120 mm Hg. These cardiovascular changes were accompanied by wide dilatation of the pupils and brisk retraction of the nictitating membranes. In about one-quarter of the animals piloerection occurred and in a smaller number jaw movements were also seen. Piloerection and jaw movements were more readily elicited if the stimulus strength was increased but for all except the last few tests in each experiment the stimulating current was deliberately held

as low as was consistent with an adequate cardiovascular response, both to minimize the possibility of brain damage at the electrode tip with repeated excitation and also to avoid evoking limb muscle movements which would lead to secondary cardiovascular adaptations.

The measured reduction in hind limb perfusion pressure probably underestimated the dilatation in the muscle bed since it was not always possible to completely exclude collateral flow. Collateral flow was tested for by

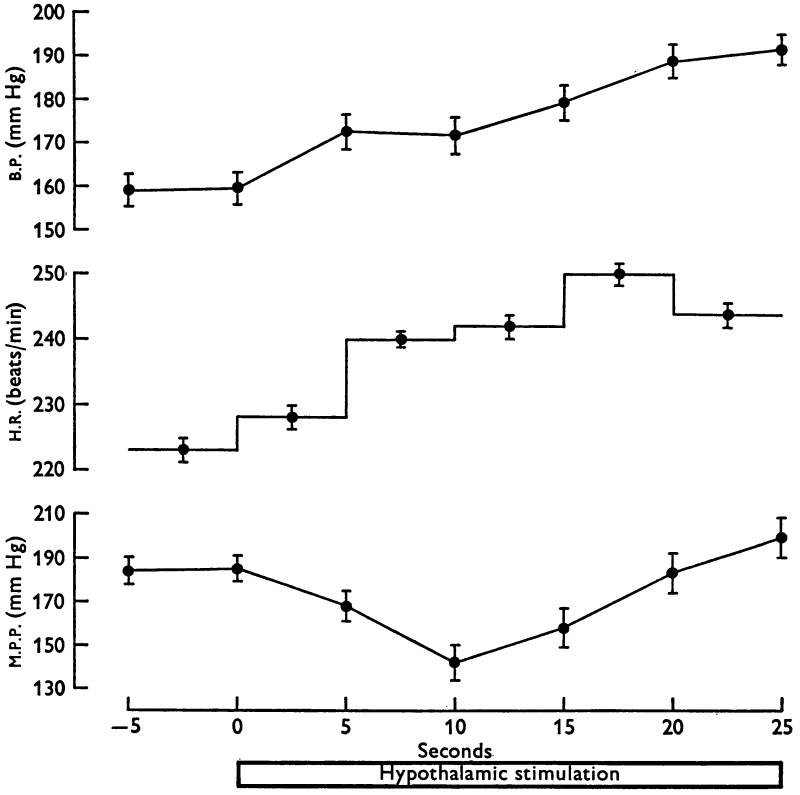

Fig. 2. Mean values (± S.E.) of aortic blood pressure (B.P.), heart rate (H.R.) and inflow pressure to the skinned hind limb perfused at constant flow (M.P.P.) during twenty-six trials in fifteen cats of a 25 sec period of hypothalamic stimulation. The values for B.P. and M.P.P. were measured at 5 sec intervals; H.R. values were measured over successive 5 sec periods. One carotid sinus was denervated and the other maintained throughout at a constant pressure, the mean value of which was 93 mm Hg.

switching off the pump and observing the extent of the reduction in pressure in the perfusion circuit. Though the pressure generally fell to less than 30 mm Hg we felt that if it was below 50 mm Hg, vascular isolation,

even though possibly incomplete, was adequate, since we required only a qualitative indication of muscle vasodilatation to confirm that the electrode had been positioned in a suitable hypothalamic site capable of evoking activity of the sympathetic vasodilator system.

At the end of the experiment the decrease in hind limb perfusion pressure was shown to result from activity in cholinergic vasodilator fibres by repeating the hypothalamic stimulation after the administration of atropine. However, before atropine was given the aortic nerves, if not already divided, were cut and a further trial of hypothalamic stimulation carried out. Persistence of the vasodilatation confirmed that this could not be due to a baroreceptor reflex in response to the raised systemic pressure,

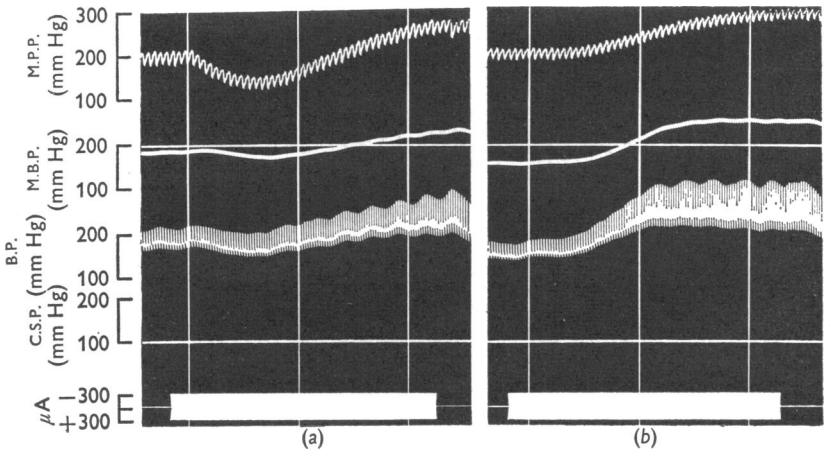

Fig. 3. Changes in pulsatile and mean aortic pressure and in muscle perfusion pressure during a 25 sec stimulation of the hypothalamus as indicated by the signal: (*a*) before atropine was given, (*b*) after atropine 0·2 mg/kg. The right carotid sinus pressure (C.S.P.) was controlled at 100 mm Hg. Both aortic nerves and the right sinus nerve were cut previously. Time marker at 10 sec intervals.

since with both aortic nerves and one sinus nerve cut, and with the pressure within the innervated, isolated sinus held constant, the baroreceptors were unlikely to be able to engender any appreciable vasodilatation. Atropine 0·2 mg/kg was then injected and the stimulation repeated. In all the seventeen cats in which this test was performed the decrease in hind limb perfusion pressure was abolished or converted to an increase with a correspondingly enhanced rise of systemic pressure. An example of such a test is shown in Fig. 3.

Sites of hypothalamic stimulation. These lay between the anteroposterior planes A 8·5 and A 11·5 (using the co-ordinates described by Horsley &

Clarke, 1908) since preliminary experiments had suggested that this region was likely to yield responses similar to that illustrated in Fig. 1. The position of the electrode tip was histologically confirmed on twenty-one occasions. These sites are depicted in Fig. 4 which shows that they were clustered in close relation to the descending column of the fornix.

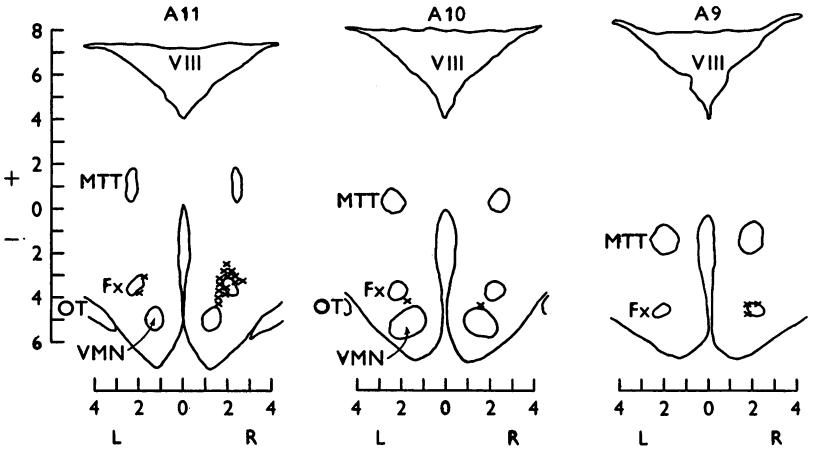

Fig. 4. Sections showing twenty-one sites of hypothalamic stimulation histologically determined in nineteen cats. Each section includes sites which lay 0·5 mm in front of and behind the plane of section. VIII, third ventricle; MTT, mamillo-thalamic tract; Fx, column of the fornix; OT, optic tract; VMN, ventro-medial nucleus. Measurements in mm.

Effects of raising the pressure within the isolated carotid sinus

The reflex response to stimulation of the carotid sinus baroreceptors was tested by abruptly raising the pressure within the vascularly isolated sinus for 15 sec. The 'low' levels of non-pulsatile sinus pressure used lay between 75 and 120 mm Hg and the 'high' levels to which sinus pressure was raised ranged from 180 to 285 mm Hg. Pressures below 75 mm Hg were never used as these are known to excite carotid body chemoreceptors (Landgren & Neil, 1951), which would complicate interpretation of the reflex cardiovascular changes. The mean changes in systemic arterial pressure, heart rate and muscle perfusion pressure (twenty-six trials in fifteen cats) are summarized in Fig. 5. All fell progressively throughout the period of raised sinus pressure though the decreases in systemic pressure and muscle perfusion pressure (43 mm Hg and 50 mm Hg respectively) were relatively more marked than the reduction in heart rate (12 beats/min).

Effect of hypothalamic stimulation on the reflex response to raised carotid sinus pressure

To determine whether the reflex response to baroreceptor stimulation was altered by simultaneous excitation of hypothalamic regions which could initiate the alerting stage of the defence response two types of experiments were performed.

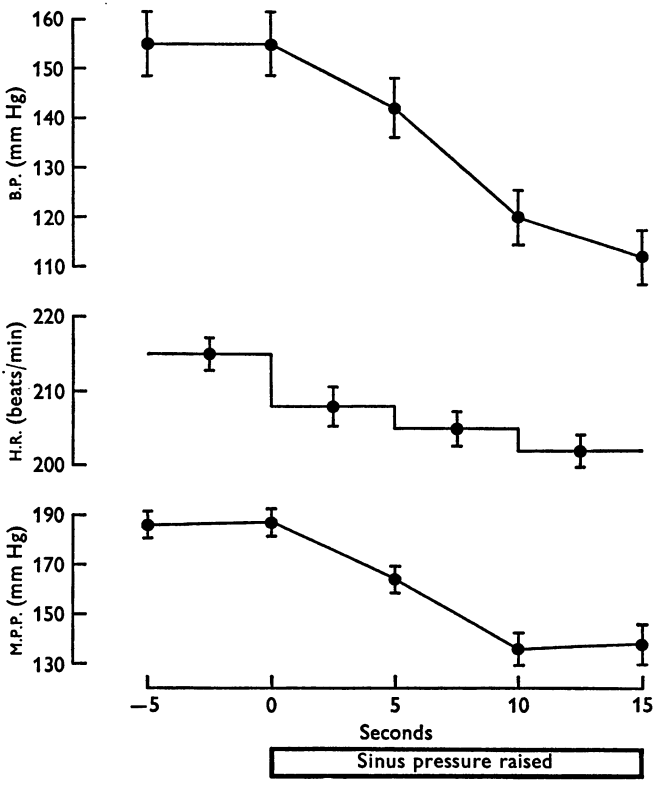

Fig. 5. The effects of raising carotid sinus pressure from a mean value of 94 (S.E. ± 2) mm Hg to 227 (S.E. ± 5) mm Hg upon systemic blood pressure (B.P.), heart rate (H.R.), and inflow pressure of a skinned hind limb perfused at constant flow rate (M.P.P.). The other carotid sinus was denervated. The results are means ± one S.E. for twenty-six trials in fifteen cats.

(*a*) Pressure within the isolated carotid sinus was increased during a period of hypothalamic stimulation. The changes in arterial blood pressure and heart rate were compared with the changes induced by a similar rise of sinus pressure but in the absence of hypothalamic stimulation.

(*b*) The effects of hypothalamic stimulation with the sinus pressure held constant at a low level were compared with the effects when sinus pressure was maintained at a high level.

(*a*) *Carotid sinus pressure raised during hypothalamic stimulation.* Each trial comprised a group of three tests; a rise in sinus pressure lasting 15 sec; a 25 sec period of hypothalamic stimulation during which the sinus pressure was held constant at its control level; and a 25 sec period of hypothalamic stimulation during which the sinus pressure was raised for 15 sec, the rise in sinus pressure either coinciding with the onset of hypothalamic stimulation or being imposed at the 10th sec. Tests were separated

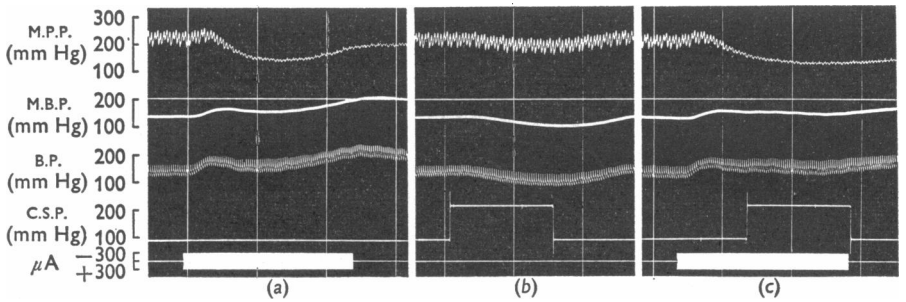

Fig. 6. Effects on mean (M.B.P.) and pulsatile (B.P.) aortic pressure and on muscle perfusion pressure (M.P.P.) of (*a*) stimulation of the hypothalamus for 25 sec with the right carotid sinus pressure (C.S.P.) controlled at 85 mm Hg; (*b*) raising the right carotid sinus pressure from 85 mm Hg to 210 mm Hg for 15 sec; (*c*) a combination of the two procedures, the sinus pressure being raised at the 10th sec of hypothalamic stimulation. The left sinus nerve was cut. Time marker at 10 sec intervals.

by an interval of at least 8 min to allow adequate time for recovery and the order of the tests was varied in different experiments. Records from an experiment of this type, in which sinus pressure was increased at the 10th sec of hypothalamic stimulation, are shown in Fig. 6.

Inspection of Fig. 6(*c*), which illustrates the effects of an increase in isolated carotid sinus pressure imposed at the 10th sec of hypothalamic stimulation, might suggest that the baroreceptor reflex was no longer operative in this situation since the fall in arterial blood pressure seen in Fig. 6(*b*), when sinus pressure was raised in the absence of hypothalamic stimulation, was apparently no longer produced. However, reference to Fig. 6(*a*) in which sinus pressure was maintained at its lower level throughout the period of hypothalamic stimulation, reveals that there would have been a continued rise of systemic pressure during the last 15 sec of stimulation were it not for the increase in sinus pressure. Thus the baroreceptor reflex effect on blood pressure remained operative, the steady level of

systemic pressure resulting from a balance of opposing hypothalamic and reflex baroreceptor influences.

The reflex effect on blood pressure elicited from the baroreceptors during hypothalamic stimulation was assessed quantitatively from the difference between the values of mean arterial blood pressure when hypothalamic stimulation was carried out with the sinus pressure maintained throughout at its control level, and the values at corresponding times when sinus pressure was raised either at the onset or at the 10th sec of stimulation. The magnitude of the response evoked by an increase in pressure in the perfused sinus during hypothalamic stimulation was then compared with the control response to the same rise of sinus pressure.

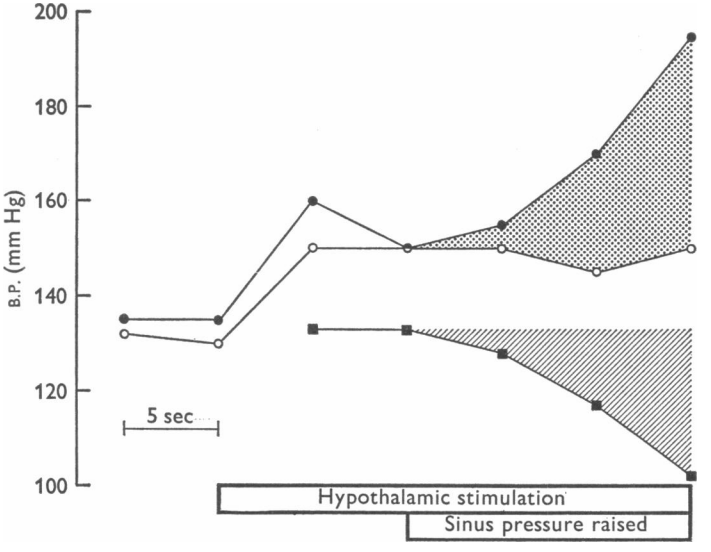

Fig. 7. Data obtained from Fig. 6 plotted to show the method of assessing the size of the reflex blood pressure response to raising carotid sinus pressure. ●, hypothalamus stimulated for 25 sec; ■, sinus pressure raised for 15 sec; ○, sinus pressure raised for 15 sec, the onset coinciding with the 10th sec of hypothalamic stimulation. Control response to elevation of sinus pressure is given by shaded area; stippled area represents response to same rise in pressure during hypothalamic stimulation.

This method of analysis is illustrated in Fig. 7 in which the blood pressure values derived from Fig. 6 have been plotted at 5 sec intervals. The stippled area between the two records for hypothalamic stimulation represents the difference due to raising the sinus pressure. The reduction in systemic pressure can be seen to be almost identical to that obtained from the sinus when the hypothalamus was not stimulated and which is represented by the shaded area. Trials of this type were performed in

twenty-three cats; in seventeen trials the sinus pressure rise was timed to coincide with the onset of hypothalamic stimulation and in forty-seven trials it occurred at the 10th sec. The mean values for systemic blood pressure and the changes reflexly induced from the isolated carotid sinus in the absence of hypothalamic stimulation and during such stimulation are illustrated in Fig. 8(*a*) and (*b*), for experiments in which the sinus pressure was raised at the onset and at the 10th sec of hypothalamic stimulation respectively. This Figure shows that the changes in pressure evoked from the sinus were virtually the same, indicating that excitation of the hypothalamus did not reduce the ability of the sinus baroreceptors

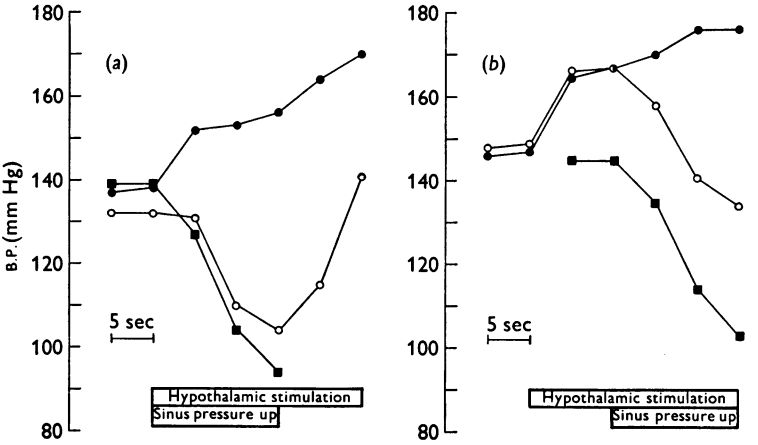

Fig. 8. The effects on mean blood pressure of stimulating the hypothalamus for 25 sec, ●; raising carotid sinus pressure for 15 sec, ■; and of the simultaneous application of these stimuli, ○. Part (*a*): results of seventeen sets of trials in twelve cats in which the right carotid sinus pressure was raised from a mean control level of 94 mm Hg to a mean level of 205 mm Hg from the onset until the 15th sec of hypothalamic stimulation. Part (*b*): results of forty-seven sets of trials in twenty-three cats in which the right carotid sinus pressure was raised from a mean control level of 87 mm Hg to a mean level of 216 mm Hg at the 10th sec of hypothalamic stimulation.

to lower the systemic pressure. Table 1 lists the mean differences between the vasodepressor responses produced by the sinus in individual paired tests with and without hypothalamic stimulation and shows that they were small, the differences generally being less than 10 % of the control response, and with one possible exception not statistically significant.

The effect of the hypothalamus on the changes in *heart rate* induced by baroreceptor stimulation was less easy to assess, since in the majority of cats raising the carotid sinus pressure led to negligible changes in heart rate. However, in seven animals heart rate fell by 20 beats or more, and

there were eighteen trials in which sinus pressure was raised during hypothalamic stimulation in this group of cats. In eight of these trials the bradycardia previously elicited from the sinus was completely suppressed during hypothalamic stimulation and in another two trials it was partially suppressed. Four of these instances of complete suppression of bradycardia and one of partial suppression were in trials in which sinus pressure was raised at the onset of hypothalamic stimulation. In the other four instances of complete suppression and one of partial suppression of reflex

TABLE 1. Effect of hypothalamic stimulation on the fall in systemic blood pressure reflexly evoked by a rise in carotid sinus pressure (C.S.P.). In deriving the effect of the rise in sinus pressure during hypothalamic stimulation any change in systemic pressure during an equivalent period of hypothalamic stimulation alone has been taken into account as indicated in Fig. 7. 'Pressure difference' is the fall in systemic pressure at the stated time after C.S.P. was raised in the absence of hypothalamic stimulation, minus the fall in systemic pressure at the same interval after C.S.P. was raised during hypothalamic stimulation. *P* is the probability that this value is significantly different from zero

A. Sinus pressure raised at onset of hypothalamic stimulation from a mean of 94 mm Hg to a mean of 205 mm Hg

Sec after onset of hypothalamic stimulation	Sec after C.S.P. raised	Number of trials	Pressure difference (mm Hg)	*P*
5	5	17	+3	$\approx 0{\cdot}4$
10	10	17	+2	$\approx 0{\cdot}4$
15	15	17	+2	$\approx 0{\cdot}4$

B. Sinus pressure raised at the 10th second of hypothalamic stimulation from a mean of 87 mm Hg to a mean of 216 mm Hg

Sec after onset of hypothalamic stimulation	Sec after C.S.P. raised	Number of trials	Pressure difference mm Hg	*P*
15	5	47	+3	$0{\cdot}05 > P > 0{\cdot}02$
20	10	47	+4	$\approx 0{\cdot}1$
25	15	42	+4	$\approx 0{\cdot}1$

bradycardia, sinus pressure was raised at the tenth second. The results of one of these experiments in which hypothalamic stimulation largely suppressed the bradycardia, even though the hypotensive response to the elevation of sinus pressure persisted, are plotted in Fig. 9. In the absence of hypothalamic stimulation raising the isolated carotid sinus pressure from 92 mm Hg to 200 mm Hg for 15 sec led to a fall in systemic B.P. of 45 mm Hg and a reduction in heart rate of 66 beats/min. During hypothalamic stimulation the same rise in sinus pressure produced an almost identical fall in blood pressure of 44 mm Hg (after making allowance, as

previously described, for the continued rise in pressure which would have occurred had sinus pressure remained steady) but the reduction in heart rate was now merely 16 beats/min.

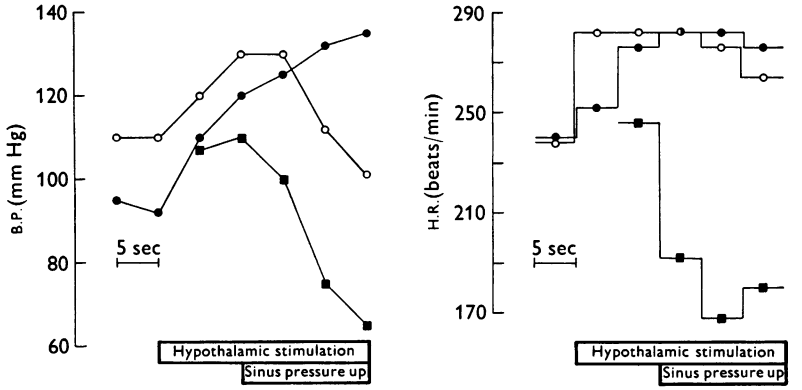

Fig. 9. Comparison of the effects of hypothalamic stimulation on reflex blood pressure and heart rate responses to a rise in sinus pressure from 92 mm Hg to 200 mm Hg. ●, hypothalamus stimulated only; ■, sinus pressure raised only; ○, sinus pressure raised at 10th sec of hypothalamic stimulation. Note persistence of reflex effect on blood pressure with almost complete suppression of reflex bradycardia.

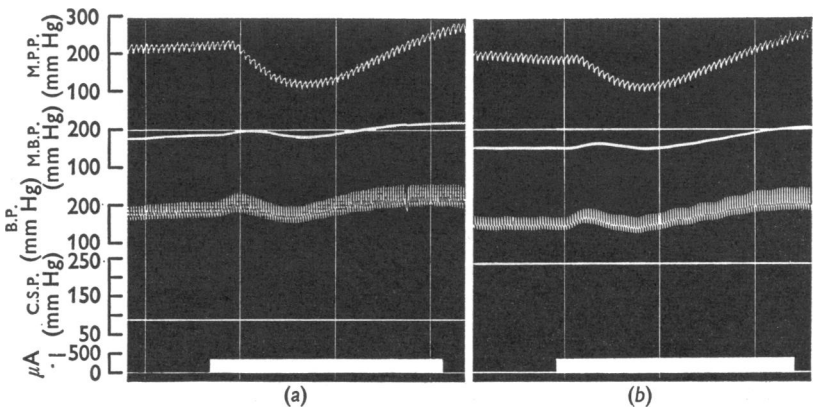

Fig. 10. Changes in mean (M.B.P.) and pulsatile (B.P.) aortic pressure and in muscle perfusion pressure (M.P.P.) during a 25 sec stimulation of the hypothalamus with the right carotid sinus pressure (C.S.P.) controlled in (*a*) at 95 mm Hg, and in (*b*) at 242 mm Hg. The left sinus nerve was cut. Time marker at 10 sec intervals.

(*b*) *Carotid sinus pressure maintained at either low or high level throughout.* Fifteen trials of this type were performed in ten cats. Each trial comprised two tests; in one the pressure in the isolated carotid sinus was held constant

at a low level lying between 80 and 120 mm Hg throughout both the control period and the period of hypothalamic stimulation; in the other the sinus pressure was held constant at a higher level, between 180 and 250 mm Hg. The records of such a pair of tests are shown in Fig. 10. The control arterial blood pressure was less in the trial with the pressure in the isolated sinus held at the higher level (Fig. 6(*b*)) because of the greater baroreceptor discharge. During hypothalamic stimulation systemic blood pressure rose in both tests but as the rise in pressure was approximately the same in each instance the difference between the systemic pressure levels reached

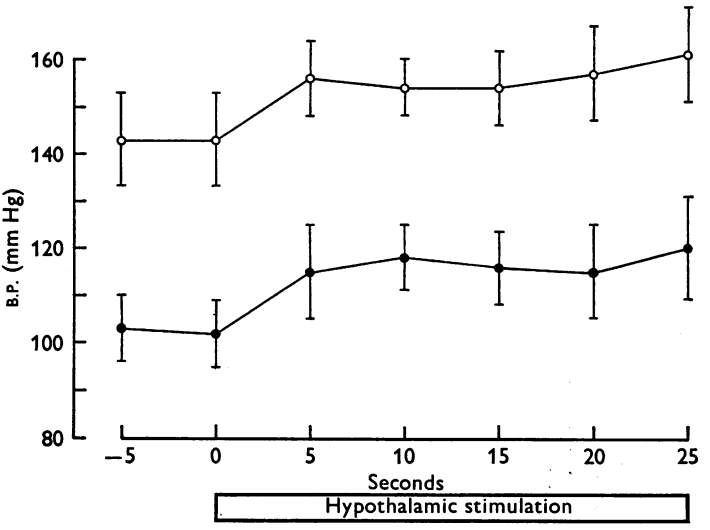

Fig. 11. Systemic blood pressure changes during paired tests of hypothalamic stimulation. In one test of each pair (○) the pressure within the carotid sinus was maintained at a steady low level (mean value 101 mm Hg); in the other test (●) the sinus pressure was maintained at a higher steady level (mean value 206 mm Hg). The other carotid sinus was denervated. The blood pressure was measured at 5 sec intervals; mean values ± 1 S.E. are shown.

during hypothalamic stimulation remained much the same as the difference present during the preceding control period. This is further exemplified by Fig. 11 in which the mean values for all the tests at low and high pressures have been plotted; the two curves run virtually parallel to one another.

The results were further analysed by comparing for each pair of tests the difference between the arterial pressure values during their control periods with the corresponding differences at the 5th, 10th, 15th, 20th and

25th sec of hypothalamic stimulation. The difference never varied by more than 4 mm Hg and in no case was the variation significant, $P > 0{\cdot}2$ in every instance.

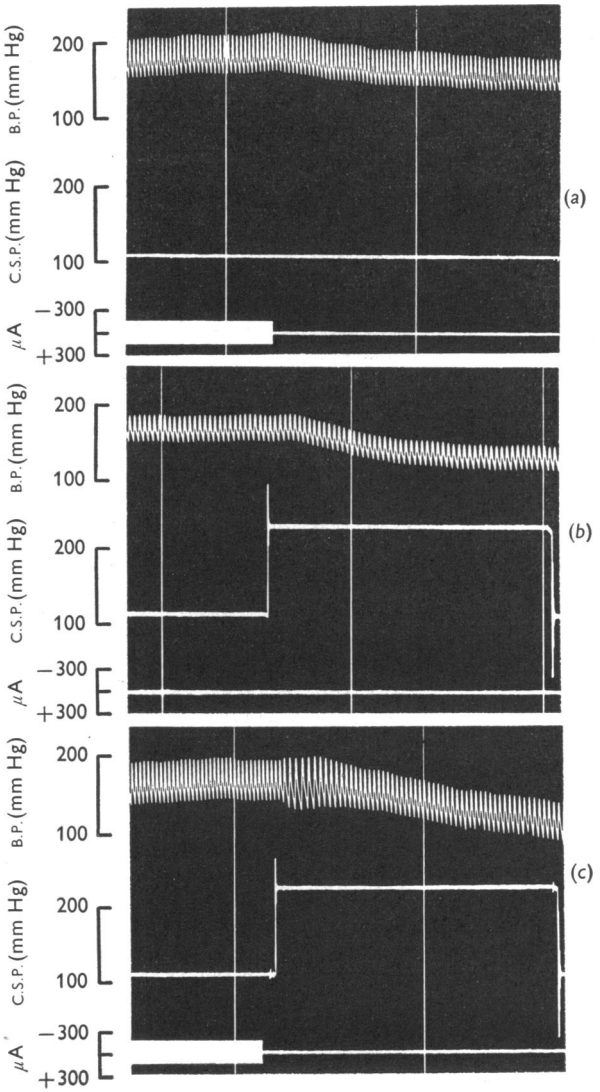

Fig. 12. Changes in aortic pressure (B.P.); (*a*) at the end of a 25 sec stimulation of the hypothalamus with the right carotid sinus pressure (C.S.P.) controlled at 108 mm Hg; (*b*) during a 15 sec period over which right carotid sinus pressure was increased from 108 mm Hg to 230 mm Hg; (*c*) during a 15 sec period of increased carotid sinus pressure beginning at the end of a 25 sec hypothalamic stimulation. Time marker at 10 sec intervals. Note bradycardia during raised sinus pressure in (*c*).

Reflex responses of arterial blood pressure and heart rate to a rise in sinus pressure immediately following hypothalamic stimulation

In seven cats, the reflex cardiovascular effects of increasing the pressure in the isolated carotid sinus were examined immediately following stimulation of the hypothalamus. The sinus pressure was maintained constant at a low level throughout the 25 sec period of hypothalamic stimulation and then abruptly raised simultaneously with the end of the stimulus. In five of these seven animals the vasodepressor response to carotid sinus stimulation appeared to be markedly delayed and reduced in that the decline in systemic pressure which normally followed the termination of hypothalamic stimulation was very little greater, at least for the first 5–10 sec, when accompanied by a rise in sinus pressure.

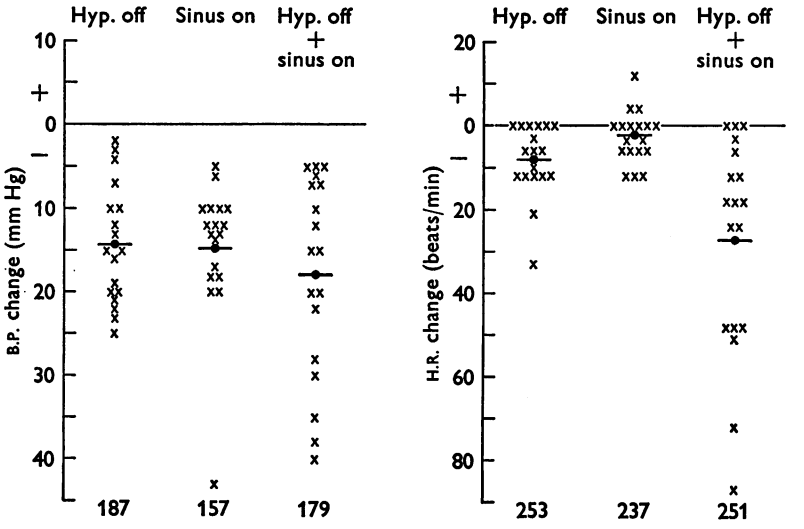

Fig. 13. Changes in mean blood pressure and heart rate during the first 5 sec following the end of 25 sec hypothalamic stimulation (hyp. off); during the first 5 sec of raised sinus pressure (sinus on) and during the first 5 sec of raised sinus pressure when the pressure rise coincided with the end of 25 sec hypothalamic stimulation (hyp. off + sinus on). Results are from eighteen trials in seven cats. Mean values are shown —●—. The numbers beneath the columns are the mean values of blood pressure or heart rate at the beginning of the corresponding 5 sec period.

Whereas the tendency was for a reduction in the hypotensive response to a rise in sinus pressure which immediately followed hypothalamic stimulation, the reflex bradycardia tended to be enhanced. An example of this is seen in Fig. 12. In this experiment neither the ending of hypothalamic stimulation (Fig. 12(*a*)) nor the application of increased pressure

to the sinus (Fig. 12(*b*)) led to any perceptible change in heart rate, but when the increase in sinus pressure followed immediately on the end of hypothalamic stimulation a very obvious bradycardia occurred (Fig. 12(*c*)).

The difference between the effects of prior hypothalamic stimulation on the reflex response of blood pressure and of heart rate to an increase in sinus pressure is further illustrated by Fig. 13 which records the results of the eighteen trials on the seven cats in this group. The Figure depicts the changes in blood pressure and heart rate during the first 5 sec after the end of hypothalamic stimulation (hyp. off), during the first 5 sec of raised sinus pressure (sinus on), and during the first 5 sec of raised sinus pressure when this immediately followed the end of hypothalamic stimulation (hyp. off + sinus on). The mean fall in blood pressure when the two procedures were combined was 17·8 mm Hg; scarcely more than the mean falls following hypothalamic stimulation or on increasing sinus pressure, 14·3 mm Hg and 14·6 mm Hg respectively. However, the equivalent mean reduction in heart rate, 27·1 beats/min, greatly exceeded even the sum of the mean reductions in heart rate following hypothalamic stimulation, 8 beats/min, and on increasing sinus pressure, 2·3 beats/min.

DISCUSSION

Several groups of workers have concluded that baroreceptor reflexes persist during hypothalamic stimulation, as indicated by the reduction in sympathetic discharge (Pitts, Larrabee & Bronk, 1941), reflex bradycardia and fall in blood pressure (Wilson, Clarke, Smith & Rushmer, 1961; Reis & Cuénod, 1965) in response to carotid sinus baroreceptor stimulation. However, as measurements of muscle blood flow were not made in these investigations there is no certainty that hypothalamic areas integrating the defence response were being studied, though in some instances the anatomical description of the position of the electrode tip suggests that this may have been so.

The areas of the hypothalamus stimulated in the present study all lay within the region found by Eliasson, Folkow, Lindgren & Uvnäs (1951) to activate a sympathetic vasodilator supply to skeletal muscle and which was subsequently shown by Abrahams *et al.* (1960) to be coextensive with the region from which Hess & Brügger (1943) had been able to evoke the defence reaction in the conscious animal. In addition to atropine-sensitive muscle vasodilatation, which was regarded as an essential confirmation of correct positioning of the electrode, we also observed the increases in blood pressure and heart rate, pupillary dilatation, retraction of the nictitating membranes and in some instances piloerection and somatic movements described by these previous workers.

Our results demonstrate that both during and immediately following stimulation of this specific hypothalamic area there are also changes in the pattern of response to stimulation of the carotid sinus baroreceptors. The most prominent reflex effects of an increase in carotid sinus pressure are a fall in blood pressure and bradycardia, though in the cat bradycardia is less marked than in other species such as the dog, and even in the absence of hypothalamic stimulation occurred in only a proportion of our experiments. However, in about half the cats in which a bradycardia could be evoked from the sinus in the present series, no bradycardia occurred if the hypothalamus was being stimulated at the same time. Similar suppression of baroreceptor induced bradycardia during stimulation of the hypothalamic defence area has been reported by Hilton (1963, 1965) and by Djojosugito, Folkow, Kylstra, Lisander & Tuttle (1970). By contrast with this suppression of the reflex bradycardia we found that the hypotensive response to carotid sinus distension was undiminished during hypothalamic stimulation. Persistence of this reflex effect on blood pressure was also illustrated by the experiments in which sinus pressure was held constant at either a low or a high level throughout both the control and stimulation periods. In these experiments the *rise* in systemic blood pressure during hypothalamic stimulation was virtually identical whether the carotid sinus pressure was being held at a low or a high level. This would not have been so if the reflex hypotensive response to baroreceptor stimulation had been suppressed during hypothalamic stimulation.

The current intensities used to stimulate the hypothalamus in this study were generally between 100 and 300 μA. Such intensities produced well marked cardiovascular responses which remained reproducible even after ten or more periods of stimulation. Djojosugito *et al.* (1970), who measured flow resistance in the muscle, skin and intestinal vascular beds in addition to recording systemic arterial pressure, similarly found that the fall in blood pressure and also the reduction of regional vascular resistance on raising carotid sinus pressure persisted during hypothalamic stimulation of comparable intensity. On the other hand, Hilton (1963, 1965) reported that stimulation of the hypothalamus for 10 sec beforehand may almost completely suppress the reflex fall in blood pressure as well as the reflex bradycardia which follow a sudden increase in pressure from 0 to 200 mm Hg within a blind sac preparation of the carotid bifurcation. Djojosugito *et al.* (1970) were able to suppress the fall in systemic pressure evoked by stimulation of the central end of the left vagus nerve when using higher intensities of hypothalamic stimulation which would appear to have been in the region of 500 μA. However, even in the presence of these more powerful stimuli there was little difference in their experiments in the extent of baroreceptor inhibition of vasomotor tone as indicated by the resistance

changes in the various vascular beds. In our animals, which unlike those of Djojosugito *et al.* were not curarized, stimulus intensities of this order usually produced movements of the limbs and trunk with their accompanying secondary cardiovascular effects. Even when such movements did not occur the responses to these more powerful stimuli could not be adequately analysed since after only a few trials the electrode current generally became unsteady with correspondingly variable cardiovascular changes, presumably due to tissue damage at the electrode tip. However, on one occasion current strength was increased to 600 μA without any such untoward effects and even at this intensity of hypothalamic stimulation the effect of the carotid sinus on systemic pressure was undiminished.

The results of a recent investigation by Gebber & Snyder (1970) closely parallel our own observations and those of Djojosugito *et al.* (1970) when using moderate stimuli. In the spinal cat with intact vagi stimulation of the hypothalamus, at sites a little lateral and posterior to the area stimulated in this study, inhibited bradycardia evoked by norepinephrine or carotid sinus nerve stimulation. Stimulation also blocked bradycardia evoked by carotid sinus stretch in cats with the neuraxis intact. By contrast, hypothalamic stimulation failed to reduce the hypotensive response produced by this procedure, and discharges induced in the splanchnic, inferior cardiac and external carotid sympathetic nerves by hypothalamic stimulation were reduced during the pressor response evoked by norepinephrine.

Our observation that a given increase in carotid sinus pressure evoked a similar fall in blood pressure whether or not the hypothalamic defence area was being stimulated simultaneously does not, however, necessarily imply that the hypothalamus was without effect on this component of the reflex response to baroreceptor stimulation. The parallelism between the changes in mean blood pressure in the paired experiments in which pressure in the isolated sinus was held constant at low or high levels during hypothalamic stimulation suggests that activity of the hypothalamus may be 'resetting' the baroreceptor mechanism so that it now operates at a higher level of pressure. Resetting of the baroreceptor reflex has also been envisaged by Hilton (1966) though as already mentioned Hilton found that the reflex fall in blood pressure as well as the reflex bradycardia was almost completely suppressed during hypothalamic stimulation. A more specific examination of the effects of hypothalamic stimulation on the operating level and on the threshold and sensitivity of the baroreceptor reflex is obviously needed.

The results of the present study also show that the baroreceptor reflex is modified during the initial five or ten seconds immediately after hypothalmic stimulation, but now it is the effect on blood pressure which is

reduced or absent whereas the normal bradycardia may be very considerably enhanced. A similar pattern of alteration in heart rate responses to a rise in sinus pressure has been reported by Hockman, Talesnik & Livingston (1969) who found that the bradycardia induced by electrical stimulation of the sinus nerve was inhibited by simultaneous stimulation of the central grey matter of the mid-brain, but that immediately following such stimulation the bradycardia was enhanced, and this facilitatory influence persisted for up to 10 sec after the end of the stimulus. Hilton (1965) also noted that a marked bradycardia often occurred within a few seconds of stopping hypothalamic stimulation and attributed this to post-inhibitory rebound of the baroreceptor reflex. In our own experience the baroreceptor reflex responses had always returned to normal 15 sec after the end of hypothalamic stimulation. This time course of the facilitation of baroreceptor induced bradycardia and the similar time course of the inhibition of reflex vasodepression which we also observed following the end of stimulation suggest that these alterations in the reflex responses to baroreceptor stimulation may be related to the depression of sympathetic nerve impulse activity noted by Pitts *et al.* (1941) during the first few seconds following a period of hypothalamic stimulation. However, further analysis of the mechanisms responsible for the changes in the baroreceptor reflex responses during and following hypothalamic stimulation is still required.

An indication of the possible functional significance of the different effects of hypothalamic activity on the various components of the baroreceptor reflex response, and which presumably occur during the alerting phase of the defence response, has been given by Kylstra & Lisander (1970). These authors showed that persistence of the carotid sinus vasomotor reflex combined with suppression of reflex bradycardia during defence area stimulation permitted the same cardiac output to be sustained with a reduced ventricular work load, or alternatively, for the same ventricular work load a greater cardiac output was possible. Moreover, since the vasodilatation seemed to favour the muscle vasculature, the metabolically significant factor, the flow through the muscle bed, was still further enhanced. Thus, as Kylstra & Lisander stress, this altered pattern of response enables the baroreceptor reflex to compliment the defence reaction rather than oppose it.

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